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Differential chemical hazards of common insecticides to Eupelmidae parasitoids: An ecological risk assessment for integrated pest management in forest ecosystems

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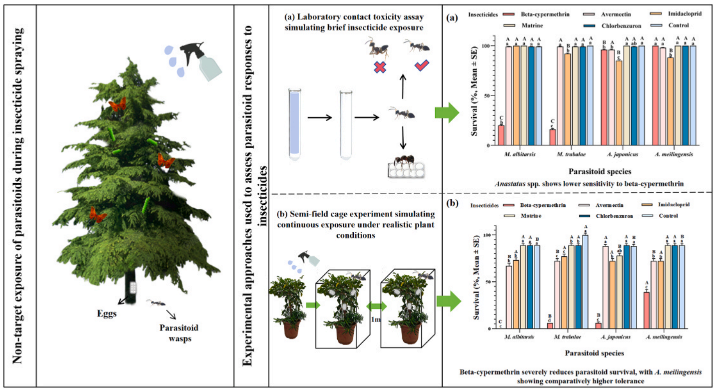
HIGHLIGHTS

- Chemical insecticides often negatively impact parasitic wasps.
- Eupelmidae is a key family of biological control agents for fruit and forest pests.
- Parasitoids of the genera *Anastatus* and *Mesocomys* each exhibit their own similar patterns of susceptibility.
- Natural high resistance to pyrethroids was first discovered in *Anastatus*.

GRAPHICAL ABSTRACT



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ABSTRACT

Eupelmidae parasitoids (*Anastatus* and *Mesocomys* spp.) are crucial for biological pest control, yet their ecological function may be compromised by insecticide exposure within integrated pest management programs. We evaluated the lethal and sublethal hazards of five widely used insecticides (beta-cypermethrin, avermectin, imidacloprid, matrine, and chlorbenzuron) on four parasitoid species. Beta-cypermethrin exhibited high toxicity to *M. albitarsis* and *M. trabalae*, causing 100 % mortality within 72 h, while *A. japonicus* and *A. meilingensis* showed significantly greater tolerance. Hazard quotient (HQ) analysis confirmed the high risk of beta-cypermethrin to *Mesocomys* species (HQ > 1), whereas *Anastatus* species (HQ < 1) were less affected. In contrast, matrine and chlorbenzuron produced minimal adverse effects on survival, parasitism, emergence, or sex ratio across all tested species. Further experiments revealed that *A. meilingensis* maintained survival rates exceeding 70 % following exposure to other pyrethroids, whereas survival of *M. albitarsis* and *M. trabalae* declined below 50 %, indicating pronounced genus-specific sensitivity to chemical stressors. Consistent patterns were observed in semi-field cage

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experiments, in which *A. meilingensis* exhibited higher survival under beta-cypermethrin exposure than the other parasitoids. Overall, the study highlights the clear contrast in insecticide tolerance between *Anastatus* and *Mesocomys* species, with *A. meilingensis* emerging as a more resilient candidate for biological control in insecticide-treated environments. Our findings support the use of selective insecticides (e.g., matrine and chlorbenzuron) to conserve vulnerable parasitoids while maintaining pest control efficacy, thereby promoting sustainable forest management.

1. Introduction

Forestry and fruit cultivation in China face serious threats from a diverse array of insect pests, with nearly 5000 species reported in forests and over 1000 species affecting fruit trees [1,2]. Major forestry pests include *Dendrolimus* spp. and *Lymantria dispar* Linnaeus, while economically important fruit pests include *Halyomorpha halys* (Stål) and *Tessaratoma papillosa* (Drury). Several species, such as *Rinaca japonica* (Moore) and *Hyphantria cunea* (Drury), attack both forest and fruit trees, resulting in substantial and recurring economic losses [1,3–5]. Given the scale, diversity, and persistence of insect pest pressures in forest and fruit production systems, long-term and environmentally compatible management strategies are urgently needed.

In this context, integrated pest management (IPM) approaches that reduce reliance on chemical insecticides have gained increasing attention. Biological control represents a core component of IPM, utilizing natural enemies to regulate pest populations while minimizing ecological disruption. The effectiveness of biological control has been demonstrated across a wide range of cropping systems, including forests, orchards, and other perennial agroecosystems [6,7]. Numerous successful applications of hymenopteran parasitoids illustrate their value within IPM programs, such as augmentative and classical biological control against *Phthorimaea absoluta* (Meyrick), *Drosophila suzukii* (Matsumura), *Pyrausta nubilalis* (Hubner), *Trialeurodes vaporariorum* (Westwood), and *T. papillosa* [8–14].

In forest and fruit ecosystems, egg parasitoids play a particularly critical role, as they suppress pest populations at the earliest developmental stage and prevent subsequent crop damage. Egg parasitoids are particularly well suited to perennial systems such as forests and orchards, where pest outbreaks are recurrent and large-scale insecticide applications can disrupt ecological balance. Consequently, egg parasitoids have become a cornerstone of biological control programs targeting lepidopteran and hemipteran pests in these systems. Within this group, parasitoids of the genera *Anastatus* and *Mesocomys* (Hymenoptera, Eupelmidae) are the most important biological control agents of forest and fruit pests. Augmentative releases of *Anastatus japonicus* Ashmead, *A. bifasciatus* (Geoffroy), *A. meilingensis* Sheng & Yu, *A. fulloi* Sheng and Wang, and *A. orientalis* Yang and Choi have effectively suppressed populations of *H. halys*, *Riptortus pedestris* (Fabricius), *T. papillosa,* and *Lycorma delicatula* (White) [4,5,15,16,17]. Similarly, several *Mesocomys* species, including *M. albitarsis* Ashmead, *M. pulchriceps* Cameron, and *M. trabalae* Yao, Yang & Zhao, are dominant egg parasitoids of major lepidopteran forest pests such as *Dendrolimus* spp., *R. japonica,* and *Imbrasia cytherea* (Fabricius) [3,18–20]. Importantly, both *Anastatus* and *Mesocomys* can be mass-reared on eggs of *Antheraea pernyi* (Guérin-Méneville) and released at large scale, enhancing their practical value in forest and orchard pest management [21].

Despite their well-documented efficacy and widespread application, chemical insecticides remain the primary tool for managing most forest and fruit pests. Increasing evidence indicates that even environmentally realistic concentrations of agrochemicals can exert pervasive sublethal effects on beneficial insects, compromising survival, behavior, and biological control services [22]. For example, pyrethroids (e.g., beta-cypermethrin), neonicotinoids (e.g., imidacloprid), macrocyclic lactones (e.g., abamectin), insect growth regulators (e.g., chlorbenzuron), and botanical insecticides (e.g., matrine) are widely used against key pests such as *Dendrolimus* spp., *R. japonica*, and *H. halys* [23,24], [25]). Although these compounds can rapidly suppress pest outbreaks, their frequent and sometimes irrational application has accelerated resistance development and increased risks to non-target beneficial insects [26,27].

In recent decades, declines in parasitic wasp abundance have been widely reported, likely driven by unregulated insecticide use and other anthropogenic factors [28–30]. During foraging and host searching, parasitoid wasps are frequently exposed to insecticide residues in their environment. Such exposure can negatively affect survival, longevity, foraging behavior, and reproductive performance, ultimately reducing biological control efficacy [22]. For example, sublethal concentrations of thiamethoxam, abamectin, and sulfoxaflor reduce longevity and control efficiency of *Eretmocerus hayati* Zolnerowich & Rose [31], while cypermethrin exposure prolongs host-searching time in *Trichogramma chilonis* Ishii [32]. Consequently, assessing the effects of insecticides on parasitoid wasps has become an important criterion for evaluating insecticide safety within IPM programs. Such assessments are essential for optimizing pest management strategies by balancing chemical control with biological regulation and reducing ecological risks [22,33]. However, despite the frequent coexistence of Eupelmidae parasitoids and insecticides in forest and orchard ecosystems, their interactions remain insufficiently characterized.

Therefore, this study aimed to evaluate the lethal and sublethal effects of five commonly used insecticides (beta-cypermethrin, imidacloprid, avermectin, matrine, and chlorbenzuron) on four key Eupelmidae parasitoids (*A. japonicus, A. meilingensis, M. albitarsis,* and *M. trabalae*), widely employed in forest and orchard pest management. In addition, the sensitivity of these four parasitoid species to other commonly applied pyrethroids (deltamethrin, bifenthrin, and cyfluthrin) was assessed to better screen for insecticide-tolerant natural enemies. Laboratory bioassays were complemented with semi-field cage experiments to enhance ecological relevance, thereby providing a scientific basis for selecting insecticides compatible with key parasitoids and optimizing integrated chemical–biological strategies for sustainable forest management.

2. Materials and methods

2.1. Insects

Cocoons of *A. pernyi* were collected in December 2023 from Yongji County, Jilin Province, China (125°48′9″ ~ 126°40′1″ E, 43°19′7″ N), and transferred to the laboratory. They were stored at 2–4℃ in darkness to maintain viability. When required, cocoons were transferred to a warm chamber (25 ± 1℃, 60 ± 10 % RH) to induce emergence of adult females. Females approaching emergence were collected daily at 8:00 a.m., and their eggs were collected for experimental use [21].

In March 2017, colonies of *A. japonicus*, *A. meilingensis*, *M. albitarsis,* and *M. trabalae* were established from wild eggs of *R. japonica* collected in an organic walnut orchard in Longnan City, Gansu Province (105°51′4″ E, 33°28′7″ N), which had not been treated with chemicals for at least five years. All species were identified based on morphological characteristics and confirmed by Dr. Gary Gibson (Agriculture and Agri-food Canada) in 2018 [3,34–36]. Laboratory trials verified that *A. pernyi* eggs were suitable hosts for rearing these parasitoids. Female moths were dissected to collect eggs, which were washed with water, air dried at room temperature, and offered to parasitoids [21]. The parasitoid colonies had been reared over more than 30 successive generations on *A. pernyi* eggs without insecticide exposure prior to testing.

2.2. Insecticides

Five insecticides commonly used against forest and fruit pests were selected for toxicity tests: beta-cypermethrin, imidacloprid, avermectin, matrine, and chlorbenzuron. These represent different classes and modes of action [37]. To further assess pyrethroid tolerance in *A. japonicus*, *A. meilingensis*, *M. albitarsis* and *M. trabalae*, three additional pyrethroids (deltamethrin, bifenthrin, and cyfluthrin) were tested (Table 1).

2.3. Contact toxicity

Each insecticide was diluted with water to its recommended field concentrations. Water was used as a control. One milliliter of each solution was added to a test tube (3 cm diameter × 12 cm height), which was gently rotated to distribute the solution evenly on the inner wall, then dried under a dark fume hood for 1 h [32]. Ten newly emerged (< 6 h old) parasitoids were placed into each treated test tube for 1 h. After exposure, parasitoids were transferred to clean tubes of the same size, provided with 20 % honey water, and maintained at 25 ± 1 °C, 70 ± 5 % RH, with a photoperiod of 14:10 h (L:D). Survival was monitored daily for 15 days. Parasitoids unable to crawl or failing to respond to gentle brush stimulation were recorded as dead. Both treatment and control groups were replicated 20 times.

2.4. Toxicity on parasitoid adults

Insecticides that caused ≥ 50 % mortality within 24 h at the recommended field concentration were selected for LC_{50} estimation. Only beta-cypermethrin met this criterion for *M. albitarsis* and *M. trabalae*. Serial dilution of beta-cypermethrin (1.64–25.43 mg a.i. L^{-1} for *M. albitarsis*; 3.05–25.43 mg a.i. L^{-1} for *M. trabalae*) were prepared based on preliminary sensitivity tests. The bioassay procedure followed the same method as described in 2.3. Survival was assessed after 24 h, with each concentration replicated 20 times.

2.5. Effects of insecticides on reproductive capacity

To evaluate the impact of insecticides on parasitism, newly emerged female parasitoids (< 6 h old) were exposed to each insecticide at field-recommended concentrations for 1 h, transferred to clean tubes, and fed 20 % honey water for 3 days. Surviving females were then given 60 fresh *A. pernyi* eggs to parasitize for 24 h. Eggs were collected, labeled, and incubated in tubes covered with white organza under controlled conditions. Emerged parasitoids, sex ratios, and number of unemerged (dead) individuals were recorded using a binocular microscope (SZ2-ILST). Both treatment and control groups were replicated 20 times.

2.6. Sensitivity of parasitoid adults to pyrethroid insecticides

To investigate broad-spectrum tolerance pyrethroids, three additional pyrethroids (deltamethrin, bifenthrin, and cyfluthrin) were tested alongside beta-cypermethrin. The protocol strictly followed the procedures described in 2.3, with survival recorded at 24 h. Both treatment and control groups were replicated 20 times.

2.7. Cage experiment

To better reflect field-relevant exposure scenarios, cage experiments were conducted to evaluate the effects of different insecticide treatments on the survival and reproductive performance of four parasitoid species. Experiments were performed in rearing cages (40 × 40 × 60 cm). Potted bougainvillea plants (*Bougainvillea spectabilis* Willd.) of uniform growth were interplanted with *Podocarpus macrophyllus* (Thunb.) Sweet to simulate the compound planting structure commonly found in forest and orchard ecosystems and to enhance ecological realism. Plants were evenly sprayed with insecticides at their recommended field concentrations and allowed to air-dry for 1 h. Subsequently, egg cards of *A. pernyi*, each containing 30 eggs, were introduced into each cage, with three cards evenly distributed on the abaxial leaf surfaces within the canopy layer. Parasitoids were released at the plant stems, mimicking standard forestry release practices, where parasitoids are typically released on tree trunks or stems and disperse upward within the canopy. All four parasitoid species were released simultaneously within the same cage, reflecting their natural co-occurrence, as these species were originally collected from walnut trees and emerged from the same host egg masses in the field [3]. This design was intended to better simulate realistic ecological conditions rather than isolate species-specific interactions. For each species, three mated females, previously fed with 20 % honey solution for three days, were released per cage. After 24 h, adult parasitoid survival was recorded, and egg cards were collected and transferred to a climatic incubator maintained at 25 ± 1 °C, 70 ± 5 % RH, with a photoperiod of 14:10 h (L:D) for development. Emergence was monitored daily, and the species and number of emerged parasitoids were recorded. If no emergence occurred for 10 consecutive days, unemerged eggs were dissected to confirm parasitoid development. Both treatment and control groups were replicated three times.

2.8. Data analyses

Data analyses were performed using IBM SPSS Statistics 20.0 (SPSS Inc., Chicago IL). LC_{50} values, regression equations, and 95 % confidence intervals were estimated with Probit analysis. The Hazard

Table 1
Formulation, field-recommended concentration, and manufacturer of the tested insecticides.

Active ingredients	Chemical classes	IRAC group	Formulation		Field-recommended concentration (mg a. i. L^{-1})	Manufacturer
			Type	% of a. i.		
Avermectin	Abamectin	6	EC	1.8	9.53	Shandong Jinnonghua Pharmaceutical Co., Ltd.
Chlorbenzuron	Benzoylurea	15	SC	25.0	167.00	Shanghai Hulian bio-pharmaceutical (Xiayi County) Co., Ltd.
Imidacloprid	Neonicotinoids	4 A	WG	70.0	46.90	Bayer crop science (China) Co., Ltd.
Beta-cypermethrin	Pyrethroids	3 A	EC	4.5	25.43	Hebei Zhongbao green crop Technology Co., Ltd
Bifenthrin	Pyrethroids	3 A	EC	10.0	33.00	Hebei Zhongbao green crop Technology Co., Ltd
Deltamethrin	Pyrethroids	3 A	EC	2.5	8.25	Bayer crop science (China) Co., Ltd.
Lambda-cyhalothrin	Pyrethroids	3 A	ME	5.0	1.65	Hebei Zhongbao green crop Technology Co., Ltd
Matrine	Quinolizidine alkaloid	-	AS	0.3	9.90	Rushan Hanwei Biotechnology Co., Ltd.

Note: "-" indicates that information could not be found in IRAC.

Quotient (HQ) was calculated as HQ = maximum field-recommended rate (g a.i. ha^{-1}) / LC_{50}. HQ > 1 was interpreted as indicating potential risk to non-target insects [38].

On the last day of observation (i.e., the 15th day), the parasitic wasps that remained alive were marked as censored data (status = 0). The survival data were analyzed using the Kaplan-Meier estimator with Log-rank tests and multiple comparison tests ($p < 0.05$). Parasitism, emergence, female offspring proportion, and 24-h survival rate were first tested for normality with Shapiro-Wilk test. When normality was not met, generalized linear models (GLMs) were applied. Parasitism were analyzed using a Poisson family GLM with a logarithmic link, considering parasitoid species and insecticide treatment as factors. Emergence rates, female offspring proportion, and survival proportions were analyzed with a Binomial GLM with a logit link [39].

Model fit was assessed with goodness-of-fit tests, and the significance of main effects and interactions was evaluated with Wald's Chi-squared tests. When significant differences were found, pairwise comparisons were performed using LSD post hoc tests. Figures were prepared using GraphPad Prism 12.5 (GraphPad Software, LLC, California, USA).

3. Results

3.1. Survival of four parasitic wasps exposed to five insecticides

Survival curves of *M. albitarsis*, *M. trabalae,* and *A. japonicus* differed significantly across the six treatments, whereas *A. meilingensis* showed no significant differences among treatments (Log-rank tests: *M. albitarsis*: $\chi^2 = 54.3$, $df = 5$, $p < 0.001$; *M. trabalae*: $\chi^2 = 68.6$, $df = 5$, $p < 0.001$; *A. japonicus*: $\chi^2 = 22.2$, $df = 5$, $p < 0.001$; *A. meilingensis*: $\chi^2 = 6.2$, $df = 5$, $p = 0.289$, Fig. 1). For *M. albitarsis*, beta-cypermethrin significantly reduced survival compared to all other treatments (Avermectin: $\chi^2 = 17.1$, $p < 0.001$; Imidacloprid: $\chi^2 = 17.1$, $p < 0.001$; Matrine: $\chi^2 = 21.1$, $p < 0.001$; Chlorbenzuron: $\chi^2 = 21.1$, $p < 0.001$; Control: $\chi^2 = 21.1$, $p < 0.001$, Fig. 1A). No significant differences were observed among the other treatments. Similarly, *M. trabalae* showed complete mortality by day two under beta-cypermethrin, which differed significantly from other treatments (Avermectin: $\chi^2 = 18.6$, $p < 0.001$; Imidacloprid: $\chi^2 = 16.8$, $p < 0.001$; Matrine: $\chi^2 = 21.2$, $p < 0.001$; Chlorbenzuron: $\chi^2 = 21.2$, $p < 0.001$; Control: $\chi^2 = 22.5$, $p < 0.001$, Fig. 1B). Under abamectin, survival was significantly lowered to matrine, chlorbenzuron, and the control (Matrine: $\chi^2 = 4.3$, $p < 0.001$; Chlorbenzuron: $\chi^2 = 7.1$, $p < 0.001$; Control: $\chi^2 = 10.0$, $p < 0.001$). In *A. japonicus*, survival under beta-cypermethrin was significantly reduced compared to avermectin, matrine, chlorbenzuron, and contro (Avermectin: $\chi^2 = 4.4$, $p = 0.036$; Matrine: $\chi^2 = 11.2$, $p < 0.001$; Chlorbenzuron: $\chi^2 = 8.6$, $p = 0.003$; Control: $\chi^2 = 13.1$, $p < 0.001$, Fig. 1C). Imidacloprid also significantly reduced survival compared to control (Control: $\chi^2 = 4.1$, $p = 0.042$).

3.2. Toxicity on parasitoid adults

Mortality from avermectin, imidacloprid, matrine, and

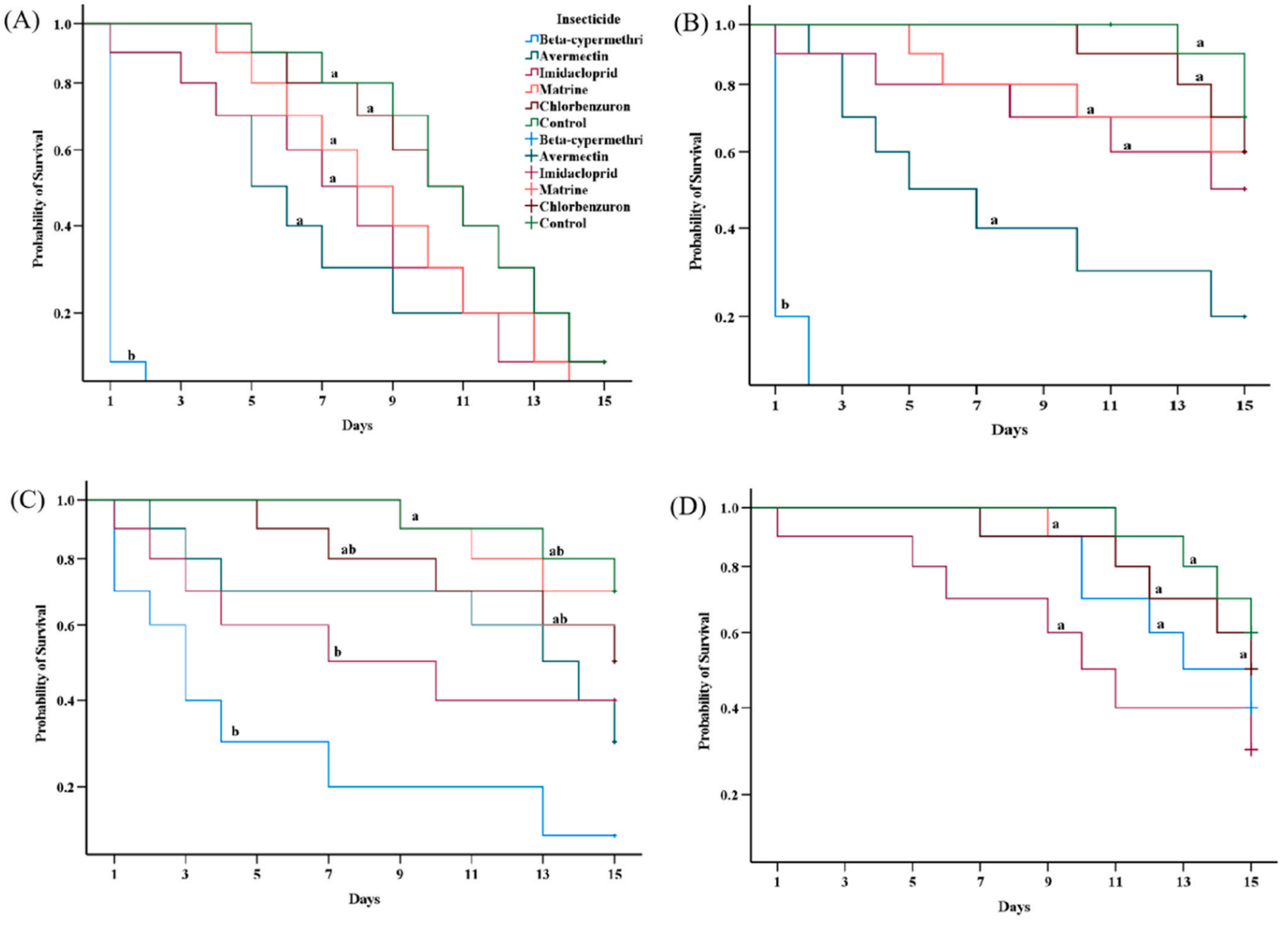

Fig. 1. Kaplan-Meier 15-days survival estimators of *Mesocomys albitarsis* (A), *Mesocomys trabalae* (B), *Anastatus japonicus* (C) and *Anastatus meilingensis* (D) exposed to five insecticides at field-recommended concentrations under laboratory conditions. To compare survival probabilities, Kaplan–Meier curves were tested using a log-rank test (5 % significance). Different lowercase letters represent the differences among various treatments.

chlorbenzuron ranged from 0 to 14.91 %. Beta-cypermethrin caused mortality of 27.97 % in *A. japonicus* and 0 % in *A. meilingensis,* preventing LC_{50} calculations for these species (Fig. 1). For insecticide–parasitoid combinations in which mortality did not reach 50 % at the recommended field concentration, LC_{50} values could not be estimated. To address this limitation, we compiled the observed mortality rates at the field-recommended concentrations and present these data in Supplementary Table S1. HQ analysis indicated that avermectin, imidacloprid, matrine, and chlorbenzuron were low risk (HQ < 1) for all parasitoids, while beta-cypermethrin was also considered low risk for *A. japonicus* and *A. meilingensis* (HQ < 1). However, for *M. albitarsis* and *M. trabalae,* beta-cypermethrin showed HQ values of 4.49 and 2.42, respectively, indicating substantial ecological risk (Table 2).

3.3. Effects of insecticides on reproductive capacity of parasitic wasps

Analysis revealed that parasitoid species, insecticide type, and their interaction significantly affected reproductive parameters including parasitism, emergence rate, and female sex ratio (Table S2).

Beta-cypermethrin exposure led to significantly fewer eggs parasitized by *A. japonicus* (14.8 eggs) and *A. meilingensis* (10.2 eggs) compared to all treatments and the control (*A. japonicus*: $\chi^2 = 109.5$, $df = 5$, $p = 0.001$; *A. meilingensis*: $\chi^2 = 150.9$, $df = 5$, $p = 0.001$; Fig. 2). No reproductive data were recorded for *M. albitarsis* and *M. trabalae* under beta-cypermethrin due to complete mortality by day 3. Other insecticides had a minimal impact on *M. albitarsis*, *A. japonicus*, and *A. meilingensis*. In contrast, *M. trabalae* parasitized significantly fewer eggs under avermectin (13.4 eggs), imidacloprid (12.8 eggs), matrine (15.1 eggs), and chlorbenzuron (12.3 eggs) than in the control (19.0 eggs) ($\chi^2 = 44.1$, $df = 4$, $p < 0.001$, Fig. 2). Overall, *M. albitarsis* and *A. japonicus* showed higher parasitism than *M. trabalae* and *A. meilingensis* under all treatments except beta-cypermethrin.

Regarding emergence (Fig. 3), *A. meilingensis* had the lowest emergence following beta-cypermethrin and matrine ($\chi^2 = 19.6$, $df = 5$, $p = 0.001$), while *M. albitarsis* had the highest emergence under avermectin, imidacloprid, and chlorbenzuron ($\chi^2 = 7.0$, $df = 4$, $p = 0.03$). However, under beta-cypermethrin treatment, *A. japonicus* emergence was significantly higher than *A. meilingensis* ($\chi^2 = 5.9$, $df = 1$, $p = 0.016$). After avermectin and matrine exposure, *A. meilingensis* emergence was significantly lower than *M. albitarsis* (Avermectin: $\chi^2 = 10.2$, $df = 3$, $p = 0.017$; Matrine: $\chi^2 = 19.0$, $df = 3$, $p < 0.001$). Under chlorbenzuron and imidacloprid, *A. japonicus* had the lowest emergence (Chlorbenzuron: $\chi^2 = 35.0$, $df = 3$, $p < 0.001$; Imidacloprid: $\chi^2 = 15.3$, $df = 3$, $p = 0.002$).

For female sex ratio (Fig. 4) *A. japonicus* and *A. meilingensis* had the lowest ratios under beta-cypermethrin ($\chi^2 = 13.6$, $df = 5$, $p = 0.019$; $\chi^2 = 49.2$, $df = 5$, $p < 0.001$), while *M. trabalae* consistently showed lower female ratios under insecticide treatments than control ($\chi^2 = 20.5$, $df = 4$, $p < 0.001$). No significant differences were observed in *M. albitarsis* ($\chi^2 = 4.4$, $df = 4$, $p = 0.352$). Across treatments (excluding beta-cypermethrin), *M. trabalae* maintained significantly lower female sex ratios than the other species (Avermectin: $\chi^2 = 65.4$, $df = 3$, $p < 0.001$; Imidacloprid: $\chi^2 = 23.0$, $df = 3$, $p < 0.001$; Chlorbenzuron: $\chi^2 = 56.1$, $df = 3$, $p < 0.001$; Matrine: $\chi^2 = 93.6$, $df = 3$, $p = 0.001$).

3.4. Survival of parasitic wasps to pyrethroid insecticides

Both species and the species-insecticide interaction significantly affected survival rates (Table S2).

For *A. meilingensis,* only bifenthrin reduced survival slightly to above 70 %, while survival under other pyrethroids remained at 100 % ($\chi^2 = 35.1$, $df = 4$, $p < 0.001$, Fig. 5). In contrast, both *M. albitarsis* and *M. trabalae* showed significantly lower survival under all pyrethroids compared to control, with survival rates below 50 % ($\chi^2 = 1847.4$, $df = 4$, $p < 0.001$; Fig. 5). Across all insecticides except the control, *A. meilingensis* exhibited significantly higher survival rates than the other three parasitoid species (Beta-cypermethrin: $\chi^2 = 1565.1$, $df = 3$, $p < 0.001$; Bifenthrin: $\chi^2 = 92.6$, $df = 3$, $p < 0.001$; Deltamethrin: $\chi^2 = 357.2$, $df = 3$, $p < 0.001$; Lambda-cyhalothrin: $\chi^2 = 579.1$, $df = 3$, $p < 0.001$).

3.5. Survival and parasitism of parasitic wasps in cage experiments

Both insecticides and the species-insecticide interaction significantly affected survival rates and parasitism of the wasps (Table S3).

Across the four parasitoid species tested in different insecticide environments, survival rates under beta-cypermethrin treatment were significantly lower than under other treatments, all falling below 50 % (*A. meilingensis*: $\chi^2 = 101.7$, $df = 5$, $p < 0.001$; *A. japonicus*: $\chi^2 = 777.2$, $df = 5$, $p < 0.001$; *M. albitarsis*: $\chi^2 = 2900.7$, $df = 5$, $p < 0.001$, *M. trabalae*:

Table 2
Acute toxicity (1-h LC_{50}) and risk analysis of five insecticides on adults of four parasitoid species.

Parasitoids	Insecticides	Regression equation	LC_{50} (95 % IF) (mg a.i L^{-1})	χ^2	HQ[a]
M. albitarsis	Beta-cypermethrin	$y = 1.574x$-1.186	5.668 (3.403–9.897)	7.975	4.49
	Avermectin	-	-	-	< 1
	Imidacloprid	-	-	-	< 1
	Matrine	-	-	-	< 1
	Chlorbenzuron	-	-	-	< 1
M. trabalae	Beta-cypermethrin	$y = 2.825x$-2.888	10.529 (7.218–16.685)	12.882	2.42
	Avermectin	-	-	-	< 1
	Imidacloprid	-	-	-	< 1
	Matrine	-	-	-	< 1
	Chlorbenzuron	-	-	-	< 1
A. japonicus	Beta-cypermethrin	-	-	-	< 1
	Avermectin	-	-	-	< 1
	Imidacloprid	-	-	-	< 1
	Matrine	-	-	-	< 1
	Chlorbenzuron	-	-	-	< 1
A. meilingensis	Beta-cypermethrin	-	-	-	< 1
	Avermectin	-	-	-	< 1
	Imidacloprid	-	-	-	< 1
	Matrine	-	-	-	< 1
	Chlorbenzuron	-	-	-	< 1

Note: "-" indicates that the mortality of parasitoids exposed to the insecticide at the recommended field concentration was less than 50 %, so LC_{50} values were not estimated. a: Hazard Quotient (HQ) = maximum field recommended rate (g a.i. ha^{-1}) / LC_{50} value estimated for the parasitoids (g a.i. ha^{-1}). HQ values > 1 indicate that insecticides have negative effects on non-target insects.

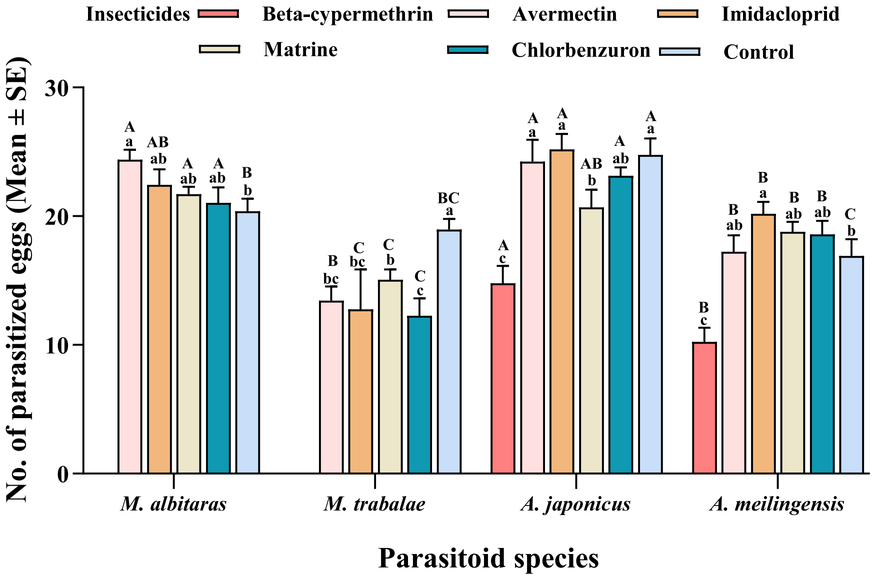

Fig. 2. Number of parasitized eggs (parasitism) by *M. albitarsis*, *M. trabalae*, *A. japonicus,* and *A. meilingensis* following exposure to five insecticides under laboratory conditions. No parasitized eggs were recorded for *M. albitarsis* and *M. trabalae* under beta-cypermethrin treatment due to 100 % mortality. Different upper-case letters indicate significant differences in the number of eggs parasitized among different parasitoid species within the same insecticide. Different lower-case letters indicate significant differences in the number of eggs parasitized by the same parasitoid species across different insecticides.

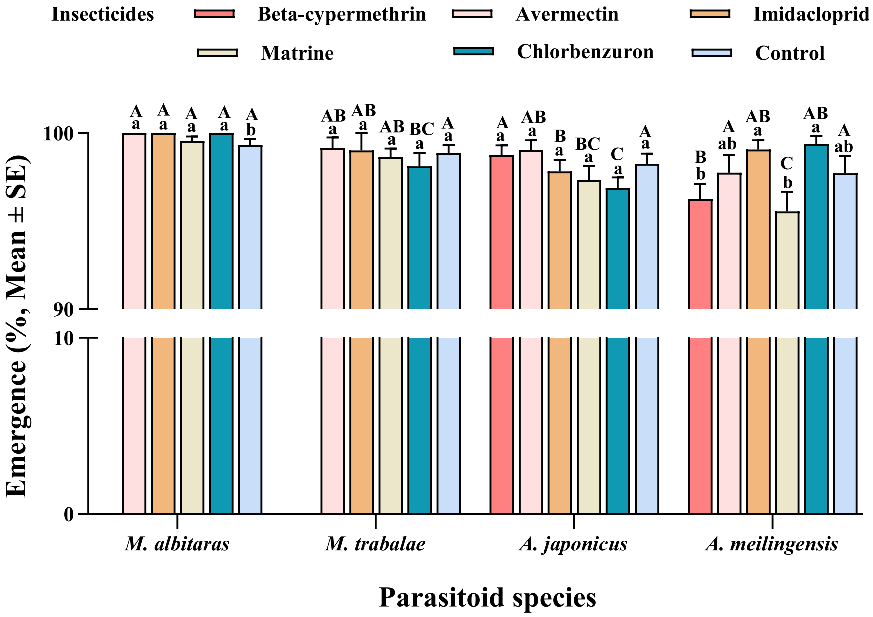

Fig. 3. Emergence rate of offspring of *M. albitarsis*, *M. trabalae*, *A. japonicus,* and *A. meilingensis* under five insecticide treatments under laboratory conditions. No emergence was recorded for *M. albitarsis* and *M. trabalae* under beta-cypermethrin treatment due to 100 % mortality. Different upper-case letters indicate significant differences in emergence rates among different parasitoid species within the same insecticide. Different lower-case letters indicate significant differences in emergence rates of the same parasitoid species across different insecticides.

$\chi^2 = 1660.1$, $df = 5$, $p < 0.001$ Fig. 6). The *A. meilingensis*, *M. albitarsis,* and *M. trabalae* all exhibited significantly lower survival rates under exposure to beta-cypermethrin, avermectin, and imidacloprid compared to the other treatments. For *A. japonicus*, apart from exhibiting the lowest survival rate under beta-cypermethrin exposure, the second lowest survival rate was observed in the imidacloprid treatment.

Under the exposure to beta-cypermethrin, the number of parasitized eggs by four species of parasitic wasps was nearly zero. In addition to beta-cypermethrin, the number of parasitized eggs by *A. meilingensis* and *A. japonicus* in imidacloprid treatments was also significantly lower than in others (*A. meilingensis*: $\chi^2 = 155.8$, $df = 5$, $p < 0.001$; *A. japonicus*: $\chi^2 = 135.0$, $df = 5$, $p < 0.001$; Fig. 7). The number of parasitized eggs by *A. meilingensis* and *A. japonicus* in environments containing avermectin, matrine, and chlorbenzuron was higher than that in the control group. The number of parasitized eggs by *M. albitarsis* under insecticide-treated conditions was significantly lower than that in the control group, while no significant difference was observed between matrine and chlorbenzuron, nor among abamectin, imidacloprid, and matrine treatments (*M. albitarsis*: $\chi^2 = 137.0$, $df = 5$, $p < 0.001$, Fig. 7). The number of parasitized eggs by *M. trabalae* in treated with beta-cypermethrin, avermectin, imidacloprid, and matrine was significantly lower than that in the chlorbenzuron and control groups, while no significant

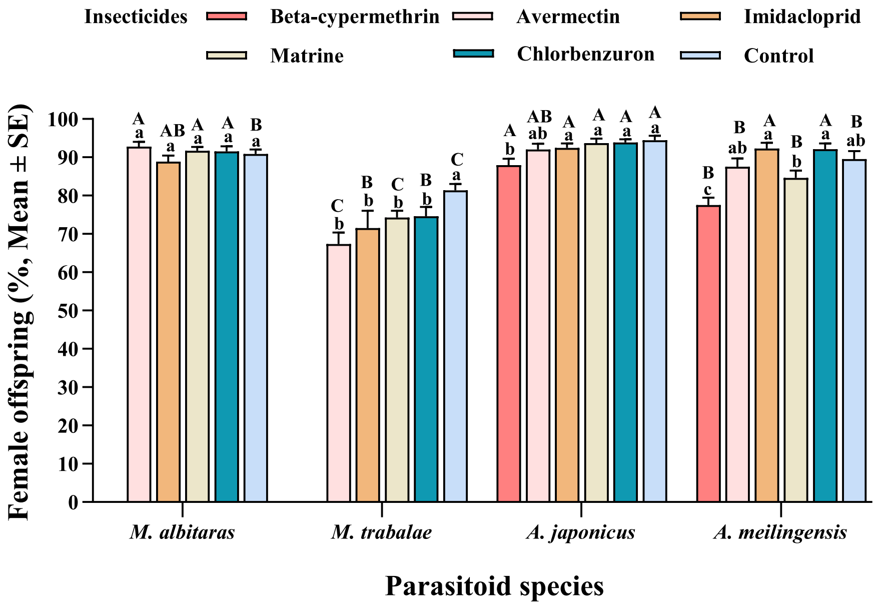

Fig. 4. Percentage of female offspring of *M. albitarsis*, *M. trabalae*, *A. japonicus,* and *A. meilingensis* under five insecticide treatments under laboratory conditions. No female offspring were recorded for *M. albitarsis* and *M. trabalae* under beta-cypermethrin treatment due to 100 % mortality. Different upper-case letters indicate significant differences female ratio among different parasitoid species within the same insecticide. Different lower-case letters indicate significant differences in female ratio of same parasitoid species across different insecticides.

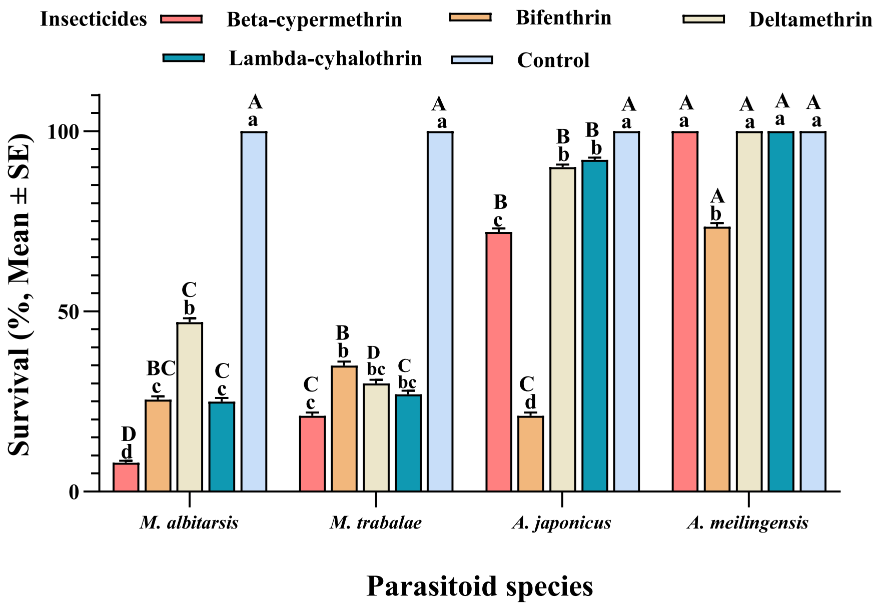

Fig. 5. Adult survival rates of *M. albitarsis*, *M. trabalae*, *A. japonicus,* and *A. meilingensis* following exposure to four pyrethroid insecticides under laboratory conditions. Different upper-case letters indicate significant differences in survival rates among different parasitoid species within the same insecticide. Different lower-case indicate significant differences in survival rates for the same parasitoid species between different insecticides.

difference was observed between the chlorbenzuron and control groups (*M. trabalae*: $\chi^2 = 58.0$, $df = 5$, $p < 0.001$, Fig. 7).

4. Discussion

Forestry and fruit industries are increasingly important, yet they face serious challenges from pest outbreaks that threaten yield and quality. In this context, parasitoids such as *Anastatus* and *Mesocomys* play a crucial role in pest suppression [34,35]. However, the widespread use of chemical insecticides for rapid pest control continues to pose significant risks to these beneficial natural enemies, as well as to the broader ecological environment [22,40]. Evaluating the safety of the commonly used insecticides for key parasitoids is therefore essential to promote sustainable pest management strategies. This study demonstrates that beta-cypermethrin exerts substantial negative effects on the four parasitoid species, while matrine and chlorbenzuron showed minimal adverse impacts. Among the tested species, *Anastatus* wasps exhibited lower sensitivity to insecticides, suggesting their suitability for large-scale release programs. These findings provide valuable insights for supporting IPM strategies in forest and fruit production systems.

In our tests, parasitoids of the same genus showed broadly similar patterns of insecticide sensitivity. Beta-cypermethrin proved highly toxic to two *Mesocomys* species, while it was less harmful to *A. japonicus* and *A. meilingensis*. Beyond beta-cypermethrin, *M. albitarsis* and

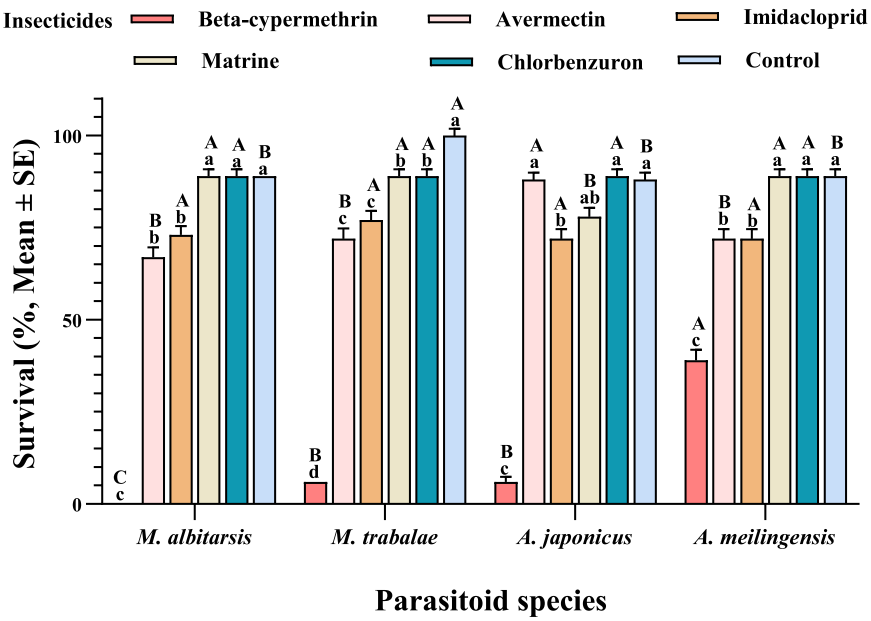

Fig. 6. Adult survival rates of *M. albitarsis*, *M. trabalae*, *A. japonicus,* and *A. meilingensis* in semi-field cage experiments following exposure to five insecticides at field-recommended concentrations. Different upper-case letters indicate significant differences in survival rates among different parasitoid species within the same insecticide. Different lower-case indicate significant differences in survival rates for the same parasitoid species between different insecticides.

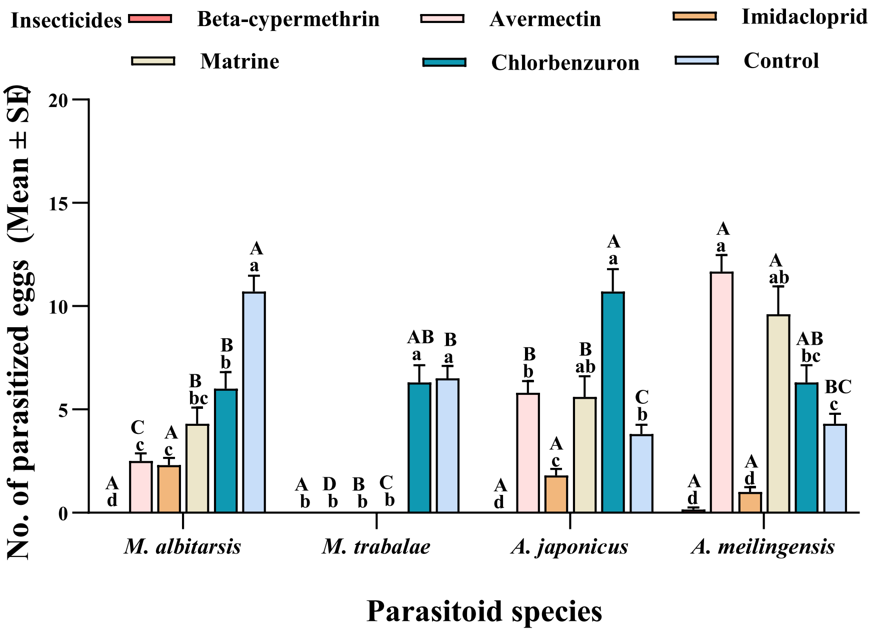

Fig. 7. Number of parasitoid eggs (parasitism) by *M. albitarsis*, *M. trabalae*, *A. japonicus,* and *A. meilingensis* in semi-field cage experiments under five insecticide treatments. Different upper-case letters indicate significant differences in the number of eggs parasitized among different parasitoid species within the same insecticide. Different lower-case letters indicate significant differences in the number of eggs parasitized by the same parasitoid species across different insecticides.

M. trabalae were also sensitive to abamectin and, to a lesser extent, imidacloprid. Cage experiments further confirmed these trends under beta-cypermethrin exposure. By comparison, *A. japonicus* and *A. meilingensis* showed greater sensitivity to imidacloprid, followed by avermectin. This difference may be related to their host ranges: *Anastatus* species parasitize a wider array of insect eggs across Hemiptera (*T. papillosa*, *H. halys* and *L. delicatula*), and Lepidoptera (*R. japonica* etc.), whereas *Mesocomys* primarily target eggs of Lepidopteran pests such as *R. japonica* and *Dendrolimus houi* Lajonquiere [3,4,11,41,42]. This broader exposure may have led *Anastatus* species to develop greater tolerance to widely used insecticides such as beta-cypermethrin and abamectin. For imidacloprid, the literature indicates it has varying degrees of activity against *Trichogramma* spp., *Aphidius gifuensis* Ashmead, and *Anagrus* sp. Chi, Su [43–45]. According to Scott [46], the P450 enzymes involved in resistance mechanisms to the same insecticide may vary among different insect populations, and multiple P450s can play dominant roles. Accordingly, the differences in insecticide susceptibility between the two genera in this study may be attributed to variation in detoxification enzyme activity, a hypothesis that warrants further physiological and molecular validation.

Beta-cypermethrin, a novel pyrethroid insecticide, is known for its neurotoxic effects, which alter the permeability of the nerve membrane and disrupt nerve conduction [47]. It is highly toxic to natural enemies. For instance, when *Eretmocerus mundus* Mercet, *Eretmocerus eremicus* Rose and Zolnerowich, and *Encarsia formosa* Gahan were exposed to synthetic pyrethrid insecticides, the mortality rate reached 100 % [48]. The current study confirmed that beta-cypermethrin was highly toxic to two *Mesocomys* species, causing 100 % mortality within three days. In contrast, it was less toxic to the two *Anastatus* species, with *A. meilingensis* exhibiting a high survival rate of 98.05 % after 3 days, while *A. japonicus* showed a lower survival rate of 35.93 % (Fig. 1). Additionally, *A. meilingensis* displayed general insensitivity to other pyrethroid insecticides (Fig. 5). Previous studies found that long-winged *Nilaparvata lugens* (Stål) were more sensitive to neonicotinoid insecticides, whereas short-winged *N. lugens* types were more sensitive to organophosphate insecticides, attributing these differences to variations in esterases and cytochrome P450 monooxygenases [49]. Li et al. [50] reported that 21 overexpressed *CarE* genes of *Plutella xylostella* (L.) populations from Hainan and Guangdong contributed to very high resistance to beta-cypermethrin. Similarly, *M. albitarsis*, *M. trabalae,* and *A. japonicus* are all long-winged parasitoids, whereas *A. meilingensis* is short-winged [3,17,51,52]. Our results indicate that long-winged parasitoids exhibit significantly higher sensitivity to pyrethroids than short-winged species. This difference may be associated with variations in detoxification enzyme expression or activity between the two morphotypes and therefore warrants further investigation. Because *A. meilingensis* is a short-winged and not-flying species, it may have evolved stronger local environmental adaptability, including tolerance to high temperatures, humidity, and chemical stresses to ensure survival [3]. Environmental regulation of juvenile hormone synthesis is known to influence wing development in insects. Under harsh conditions, long-winged morphs are favored for dispersal over long distances, whereas favorable conditions promote short-winged morphs with higher reproductive capacity, as observed for *N. lugens* and *Aphids* [53–56]. In the present study, *M. albitarsis*, *M. trabalae*, and *A. japonicus* are macropterous and capable of active flight, whereas *A. meilingensis* is brachypterous and flightless. These interspecific differences in wing morphology may reflect evolutionary trade-offs between dispersal capacity and local physiological adaptation. Based on this framework, the reduced sensitivity of *A. meilingensis* to pyrethroid insecticides is hypothesized to be associated with adaptation to local environmental stress, rather than confirmed resistance mechanisms. However, this hypothesis requires validation through targeted physiological and molecular analyses, including measurements of detoxification enzyme activities, assessments of gene expression involved in insecticide metabolism, and evaluation of potential target-site modifications. Additional contributing factors may include behavioral and genetic traits that enhance survival under chemical exposure. Previous studies have shown that species introduced through human-mediated activities may exhibit cross-resistance due to rapid adaptation to novel environments and exposure histories [57,58]. Although *A. meilingensis* is not the dominant parasitoid species on *R. japonica* eggs in Longnan City, Gansu Province, its presence in this region suggests possible human-assisted introduction followed by rapid physiological adjustment, potentially facilitated by preadaptation to plant allelochemicals [3]. Therefore, the biological performance of short-winged *A. meilingensis* under various stress conditions needs further investigation to understand its adaptive mechanisms and resilience.

In pest management, insecticides suppress pest populations mainly by disrupting physiological processes, especially reproduction, but they can also cause irreversible negative effects on the reproduction of natural enemies [22]. The results of this study showed that *M. albitarsis* and *M. trabalae* suffered 100 % mortality after exposure to beta-cypermethrin and completely lost their ability to parasitize. Although the two *Anastatus* species were still able to parasitize under beta-cypermethrin treatment, the number of parasitized eggs was significantly reduced compared to the control. Furthermore, cage experiment also demonstrated that exposure to beta-cypermethrin severely threatens the survival and reproduction of parasitic wasps. This finding is consistent with previous studies showing that pyrethroid insecticides (fenvalerate, propargite, buprofezin) affected biological parameters such as longevity, survival, fecundity, and sex ratio in *Habrobracon hebetor* (Say) [59]. Many reports suggests that reduced fertility in parasitoids can result from insecticides interfering with yolk protein synthesis [60]. This disruption affects egg formation and development, leading to egg death, mutation, or failed hatching, ultimately lowering fecundity [61]. The findings of the present study should be further validated by additional studies.

In cage experiments, the survival rates of the four parasitoid species followed trends consistent with laboratory experiments, but significant differences were observed in the number of parasitized eggs. Parasitism capacity is primarily regulated by insecticide exposure, interspecific competition, and spatial heterogeneity [22,62,63]. In the beta-cypermethrin and imidacloprid treatments, cage experiment results showed that the number of parasitized eggs by both *Mesocomys* and *Anastatus* was significantly lower than in the control group. Laboratory tests indicated that after beta-cypermethrin treatment, parasitoids of the genus *Mesocomys* nearly completely lost their parasitic ability, while *Anastatus* could still parasitize but with significantly fewer eggs compared to the control. Under imidacloprid treatment, only *M. trabalae* exhibited a significantly lower number of parasitized eggs in laboratory tests, with no significant difference observed for the other three species relative to the control. This discrepancy may stem from the parasitoids' first prolonged exposure to the insecticides, which likely impaired their host-searching and host-recognition efficiency. For instance, Bermúdez's et al. [64] found that *Tetrastichus howardi* (Olliff) avoided environments treated with spinetoram or ethiprole, hindering the parasitoids' exploration of hosts in insecticide-exposed areas. Similarly, Delpuech and Leger [65] demonstrated that *Trichogramma* wasps exposed to insecticides required more time to recognize and reject already parasitized host eggs. Under treatments with abamectin, matrine, and chlorbenzuron, the number of parasitized eggs by *Anastatus* in cage experiments was significantly higher than in the control, while no significant difference was observed in laboratory tests compared to the control. This contrast may be related to environmental complexity: test tube space is limited with no competitive pressure, whereas in the cage environment, the expanded activity range of parasitoids is accompanied by an increase in species diversity, leading to heightened interspecific competition. Additionally, prolonged insecticide exposure in cage trials may trigger adaptive behavioral adjustments in parasitoids, including even hormesis (which could enhance parasitic efficiency) [66]. The significant decline in parasitized eggs by *Mesocomys* may be due to suppressed exploratory ability, which subsequently reduces competitive pressure. In contrast, the exploratory ability of *Anastatus* is less affected, allowing it to gain an advantage in resource competition. Further experiments will be conducted to validate these findings. Interspecific competition among females may also manifest as interference or exploitation behaviors [62]. In the absence of insecticides, the number of parasitized eggs by *Mesocomys* is slightly higher than that by *Anastatus*. This may be attributed to the observation that when parasitoids of both genera encounter the same host egg patch simultaneously, *Mesocomys* exhibits significantly stronger competitive displacement ability (personal observation, unpublished data). In summary, environmental complexity (e.g., spatial scale, competitive relationships) and exposure patterns (first contact, duration) are likely key factors contributing to the discrepancies between field and laboratory results. Future research should further quantify the interactive effects of competition and insecticide stress to refine natural enemy conservation strategies and optimize integrated pest management systems. Finally, yet importantly, overparasitism or multiparasitism may have occurred (i.e., the same already-parasitized egg was parasitized again by a conspecific or heterospecific wasp). However, only one wasp can emerge from a single host egg. Therefore, the currently recorded parasitism data reflect realized successful cases, whereas the underlying processes—such as intra-egg competition—warrant further verification in future studies.

When selecting parasitoids for release, tolerance to insecticides is a critical consideration for compatibility with IPM programs. In this study, *Anastatus* species consistently exhibited higher survival and parasitism rates than *Mesocomys* species in both laboratory and semi-field trials. Accordingly, combined releases of *A. meilingensis* and *A. japonicus* may represent an effective strategy for forest and fruit pest management. It should be noted that the parasitoid populations used in this study were maintained under controlled laboratory conditions for multiple generations. Although long-term rearing ensured experimental consistency and comparability among treatments, it may have resulted in adaptation to laboratory conditions or reduced genetic variability, which could influence baseline insecticide tolerance relative to wild populations. Therefore, the present results should be interpreted primarily in a comparative context among parasitoid species under standardized conditions. Future studies incorporating recently field-collected populations will be important to confirm the ecological relevance and field applicability of these findings.

5. Conclusion

This study demonstrates that beta-cypermethrin exerts pronounced negative effects on the survival and reproductive performance of *Mesocomys* species and, to a lesser extent, on *Anastatus* species, indicating its limited compatibility with parasitoid-based biological control in forest and orchard systems. In contrast, the selective insecticides matrine and chlorbenzuron showed minimal adverse effects on all four tested parasitoid species, supporting their potential integration into IPM programs that prioritize natural enemy conservation. Although these insecticides may exhibit relatively slower or less immediate pest suppression compared with broad-spectrum pyrethroids, their selectivity enables effective pest management when applied within threshold-based decision frameworks and in combination with biological control agents. Overall, our findings highlight the greater tolerance of *Anastatus* species to chemical stress and emphasize that optimizing the balance between chemical efficacy and parasitoid conservation is essential for developing sustainable and ecologically sound pest management strategies in forestry and fruit production systems.

Environmental implication

This study reveals that widely used insecticides like beta-cypermethrin pose severe risks to *Mesocomys* parasitoids, key natural enemies of forest pests, while *Anastatus* species show higher resilience. The findings underscore the urgent need to reevaluate insecticide use in integrated pest management (IPM) to protect vulnerable parasitoids and maintain ecosystem balance. By identifying safer alternatives (matrine, chlorbenzuron), our work provides actionable solutions to reduce chemical hazards in forest ecosystems, aligning with global goals for sustainable agriculture and biodiversity conservation.

CRediT authorship contribution statement

Zhuo-Yi Zang: Writing – original draft, Methodology, Investigation, Formal analysis, Data curation. **Yong-Ming Chen:** Writing – review & editing, Resources, Funding acquisition, Formal analysis, Data curation. **Tian-Hao Li:** Writing – review & editing, Methodology, Formal analysis, Data curation. **Haneef Tariq:** Writing – review & editing, Methodology, Formal analysis, Data curation. **Antonio Biondi:** Writing – review & editing, Validation, Methodology, Formal analysis, Data curation, Conceptualization. **Jian-Fei Mei:** Writing – review & editing, Resources, Funding acquisition, Formal analysis. **Lian-Sheng Zang:** Writing – review & editing, Validation, Supervision, Resources, Project administration, Methodology, Funding acquisition, Formal analysis, Data curation, Conceptualization.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.jhazmat.2026.141413.

Data Availability

Data will be made available on request. The data that support the findings of this study are available from the corresponding author upon reasonable request.

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